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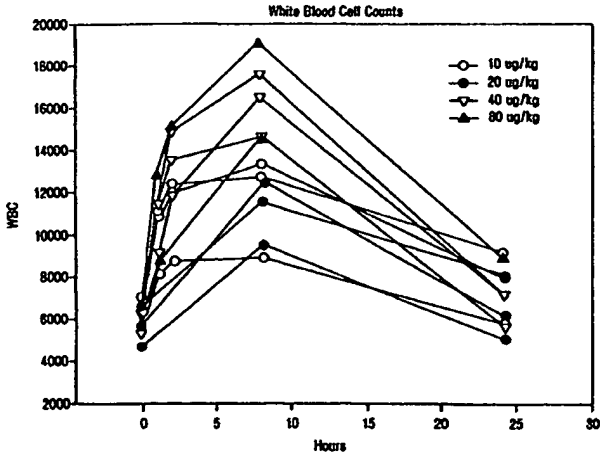

(57) Abstract

Methods to elevate white blood cell counts in animal subjects using compounds of formula (I) are disclosed. These compounds have the formula: Z–linker–Z' or pharmaceutically acceptable salt thereof wherein Z is a cyclic polyamine containing 9–32 ring members of which 3–8 are nitrogen atoms, said nitrogen atoms separated from each other by at least 2 carbon atoms, and wherein said heterocycle may optionally contain additional heteroatoms besides nitrogen and/or may be fused to an additional ring system; Z' may be embodied in a form as defined by Z above, or alternatively may be of the formula: $-N(R)-(CR_2)_n-X$ wherein each R is independently H or straight, branched or cyclic alkyl (1–6C), n is 1 or 2, and X is an aromatic ring, including heteroaromatic rings, or is a mercaptan; "linker" represents a bond, alkylene (1–6C) or may comprise aryl, fused aryl, oxygen atoms contained in an alkylene chain, or may contain keto groups or nitrogen or sulfur atoms; in an amount effective to elevate said WBC count in said subject.

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METHODS AND COMPOSITIONS TO ENHANCE WHITE BLOOD CELL COUNT

Technical Field

The invention is in the field of therapeutics and medicinal chemistry. More particularly, the invention concerns methods to enhance white blood cell counts in subjects by administering certain cyclic polyamines.

Background Art

White blood cells play a significant part in maintaining the health and viability of animals, including humans. These white blood cells include neutrophils, macrophage, and basophils/mast cells as well the B and T cells of the immune system. White blood cells are continuously replaced (as are red blood cells and clot forming cells) by the hematopoietic system in response to a number of growth factors, such as colony stimulating factors (CSF) and various cytokines. The nucleotide sequences encoding a number of these growth factors have been cloned and sequenced. Perhaps the most widely known of these is granulocyte colony stimulating factor (G-CSF) which has been approved for use in counteracting the negative effects of chemotherapy. A discussion of the hematopoietic effects of this factor can be found, for example, in U.S. Patent No. 5,582,823, incorporated in its entirety by reference herein.

While endogenous growth factors are pharmacologically effective, the well known disadvantages of employing proteins and peptides, as opposed to small molecules, as pharmaceuticals underlies the need to add to the repertoire of such growth factors compounds which are themselves small molecules. In another aspect, such small molecules are advantageous over proteins and peptides where production in large quantities are desired.

A number of cyclic polyamine antiviral agents have been described in a series of U.S. patents and applications over the last several years. These patents, U.S. Patent Nos. 5,021,409; 5,583,131; 5,698,546; and 5,817,807 are incorporated herein by

reference. Also incorporated by reference is copending application Serial No. 09/111,895 filed 8 July 1998, which describes additional compounds. These patents describe the structural characteristics of the cyclic polyamine antiviral agents.

In addition, improved methods for preparation of some of these compounds are described in U.S. Patent Nos. 5,612,478; 5,756,728; 5,801,281; and 5,606,053. The disclosures of these U.S. patents are also incorporated herein by reference in their entirety.

It has now been found that the cyclic polyamine antiviral agents described in the above-mentioned patents have the effect of enhancing production of white blood cells as well as exhibiting antiviral properties. Thus, these agents are useful where treatment affects the activities within the bone marrow resulting in leukopenia, thus controlling the side-effects of chemotherapy, radiotherapy, enhancing the success of bone marrow transplantation, enhancing wound healing and burn treatment, as well as combating bacterial infections in leukemia.

Citation of the above documents is not intended as an admission that any of the foregoing is pertinent prior art. All statements as to the date or representation as to the contents of these documents is based on the information available to the applicants and does not constitute any admission as to the correctness of the dates or contents of these documents. Further, all documents referred to throughout this application are incorporated in their entirety by reference herein.

Disclosure of the Invention

The invention is directed to methods of treating animal subjects, in particular, veterinary and human patients, who are defective in white blood cell (WBC) count, or who would benefit from elevation of WBC levels. The methods of the invention employ cyclic polyamines including those described in the patents incorporated hereinabove by reference.

In one aspect, therefore, the invention is directed to a method to elevate the white blood cells (WBC) count, in a subject in need of such WBC elevation, which

method comprises administering to said subject an amount of a compound of formula (1) or of a pharmaceutical composition thereof effective to elevate WBC levels.

In additional aspects, the invention is directed to pharmaceutical compositions containing the compound of formula (1) for use in effecting WBC count elevation in animal subject.

The compounds of formula (1) are of the formula:

Z-linker-Z' (1)

wherein Z is a cyclic polyamine containing 9-32 ring members of which 3-8 are nitrogen atoms;

said nitrogen atoms separated from each other by at least 2 carbon atoms,

wherein said heterocycle may optionally contain additional heteroatoms besides nitrogen and/or may be fused to an additional ring system.

Z' may be embodied in a form as defined by Z above, or alternatively may be of the formula

$$-N(R)-(CR_2)_n-X$$

wherein each R is independently H or straight, branched or cyclic alkyl (1-6C), n is 1 or 2, and

X is an aromatic ring, including heteroaromatic rings, or is a mercaptan;

"linker" represents a bond, alkylene (1-6C) or may comprise aryl, fused aryl, oxygen atoms contained in an alkylene chain, or may contain keto groups or nitrogen or sulfur atoms.

The preferred forms of the compounds of the invention are discussed below.

Brief Description of the Drawings

Figure 1 is a graph showing the response of individual human patients to intravenous administration of a compound of the invention.

Figure 2 is a graph showing the response in elevation of WBC counts observed in HIV-infected patients who received AMD-3100 by continuous infusion for up to 10 consecutive days.

Modes of Carrying Out the Invention

The compounds useful in the invention are of the general formula set forth as formula (1) above. Certain embodiments are preferred; included among these are the compounds set forth in the above-incorporated U.S. patents.

In general, preferred embodiments of Z and Z' are cyclic polyamine moieties having from 9-24C that include 3-5 nitrogen atoms. Particularly preferred are 1,5,9,13-tetraazacyclohexadecane; 1,5,8,11,14-pentaazacyclohexadecane; 1,4,8,11-tetraazacylotetradecane; 1,5,9-triazacylcododecane; 1,4,7,10-tetraazacyclododecane; and the like, including such cyclic polyamines which are fused to an additional aromatic or heteroaromatic rings and/or containing a heteroatom other than nitrogen incorporated in the ring. Embodiments wherein the cyclic polyamine contains a fused additional cyclic system or one or more additional heteroatoms are described in U.S. Patent No. 5,698,546 incorporated hereinabove by reference. Also preferred are

3,7,11,17-tetraazabicyclo(13.3.1)heptadeca-1(17),13,15-triene;

4,7,10,17-tetraazabicyclo(13.3.1)heptadeca-1(17),13,15-triene;

1,4,7,10-tetraazacyclotetradecane; 1,4,7-triazacyclotetradecane; and

4,7,10-triazabicyclo(13.3.1)heptadeca-1(17),13,15-triene.

When Z' is other than a cyclic polyamine as defined in Z, its preferred embodiments are set forth in U.S. Patent No. 5,817,807, also incorporated hereinabove by reference.

Preferred forms of the linker moiety include those wherein the linker is a bond, or wherein the linker includes an aromatic moiety flanked by alkylene, preferably methylene moieties. Preferred linking groups include the methylene bracketed forms of 1,3-phenylene, 2,6-pyridine, 3,5-pyridine, 2,5-thiophene, 4,4'-(2,2'-bipyrimidine); 2,9-(1,10-phenanthroline) and the like. A particularly preferred linker is 1,4-phenylene-bis-(methylene).

Particularly preferred embodiments of the compound of the formula (1) include 2,2'-bicyclam; 6,6'-bicyclam; the embodiments set forth in U.S. Patent No. 5,583,131, and in particular 1,1'-[1,4-phenylene-bis(methylene)]-bis-1,4,8,11-

tetraazacyclotetradecane, set forth in U.S. Patent No. 5,021,409, and designated herein AMD3100.

Other preferred embodiments include

N-[1,4,8,11-tetraazacyclotetradecanyl-1,4-phenylenebis(methylene)]-2-aminomethyl)pyridine;

7,7'-[1,4-phenylenebis(methylene)]bis-4,7,10,17-tetraazabicyclo-[13.3.1] heptadeca-1(17),13,15-triene;

7,7'-[1,4-phenylenebis(methylene)]bis-3,7,11,17-tetraazabicyclo[13.3.1] heptadeca-1(17),13,15-triene;

1,1'-[1,3-phenylenebis(methylene)]-bis-1,4,8,11-tetra-azacyclotetradecane;

1,1'-[1,4-phenylenebis(methylene)]-bis-1,4,8,11-tetra-azacyclotetradecane;

1,1'-[1,4-phenylene-bis-(methylene)]-bis-1,4,7,10-tetraazacyclotetradecane;

1,1'-[1,3-phenylene-bis-(methylene)]-bis-1,4,7,10-tetraazacyclotetradecane;

11,11'-(1,2-propanediyl)bis-1,4,8,11-tetraazacyclotetradecane;

N-[4-(1,4,7-triazacyclotetra-decane)-1,4-phenylenebis(methylene)]-2-(aminomethyl)pyridine;

N-[7-(4,7,10-triazabicyclo[13.3.1]heptadeca-1(17),13,15-triene)-1,4-phenylenebis(methylene)]-2-(aminomethyl)pyridine;

N-[7-(4,7,10,17-tetraazabicyclo[13.3.1]heptadeca-1(17),13,15-triene)-1,4-phenylenebis(methylene)]-2-(aminomethyl)pyridine; and

N-[4-[4,7,10,17-tetraazabicyclo[13.3.1]heptadeca-1(17),13,15-triene]-1,4-phenylenebis(methylene)]-2-(aminomethyl)pyridine.

Methods to synthesize the compounds useful in the method of the invention are set forth in the U.S. patents and application incorporated hereinabove by reference.

The compounds of the invention may be prepared in the form of prodrugs, i.e., protected forms which release the compounds of the invention after administration to the subject. Typically, the protecting groups are hydrolyzed in body fluids such as in the bloodstream thus releasing the active compound or are oxidized or reduced *in vivo* to release the active compound. A discussion of prodrugs is found in Smith and

Williams Introduction to the Principles of Drug Design, Smith, H.J.; Wright, 2nd ed., London (1988).

The compounds of the invention, as they are polyamines, may be administered prepared in the forms of their acid addition salts or metal complexes thereof. Suitable acid addition salts include salts of inorganic acids that are biocompatible, including HCl, HBr, sulfuric, phosphoric and the like, as well as organic acids such as acetic, propionic, butyric and the like, as well as acids containing more than one carboxyl group, such as oxalic, glutaric, adipic and the like. Typically, at physiological pH, the compounds of the invention will be in the forms of the acid addition salts. Particularly preferred are the hydrobromides. In addition, when prepared as purified forms, the compounds may also be crystallized as the hydrates.

The compounds of the invention may be administered as sole active ingredients, as mixtures of various compounds of formula (1), and/or in admixture with additional active ingredients that are therapeutically or nutritionally useful, such as antibiotics, vitamins, herbal extracts, antiinflammatories, glucose, antipyretics, analgesics, and the like.

The compounds of the invention may be formulated for administration to animal subject using commonly understood formulation techniques well known in the art. Formulations which are suitable for particular modes of administration and for compounds of the type represented by those of formula (1) may be found in Remington's Pharmaceutical Sciences, latest addition, Mack Publishing Company, Easton, PA.

Preferably, the compounds are administered by injection, most preferably by intravenous injection, but also by subcutaneous or intraperitoneal injection, and the like. Additional parenteral routes of administration include intramuscular and intraarticular injection. For intravenous or parenteral administration, the compounds are formulated in suitable liquid form with excipients as required. The compositions may contain liposomes or other suitable carriers. For injection intravenously, the solution is made isotonic using standard preparations such as Hank's solution.

Besides injection, other routes of administration may also be used. The compounds may be formulated into tablets, capsules, syrups, powders, or other suitable forms for administration orally. By using suitable excipients, these compounds may also be administered through the mucosa using suppositories or intranasal sprays. Transdermal administration can also be effected by using suitable penetrants and controlling the rate of release.

The formulation and route of administration chosen will be tailored to the individual subject, the nature of the condition to be treated in the subject, and generally, the judgment of the attending practitioner.

Suitable dosage ranges for the compounds of formula (1) vary according to these considerations, but in general, the compounds are administered in the range of about 0.1 μg/kg-5 mg/kg of body weight; preferably the range is about 1 μg/kg-300 μg/kg of body weight; more preferably about 10 μg/kg-100 μg/kg of body weight. For a typical 70-kg human subject, thus, the dosage range is from about 0.7 μg-350 mg; preferably about 700 μg-21 mg; most preferably about 700 μg-7 mg. Dosages may be higher when the compounds are administered orally or transdermally as compared to, for example, i.v. administration.

The compounds may be administered as a single bolus dose, a dose over time, as in i.v. or transdermal administration, or in multiple dosages.

Subjects that will respond favorably to the method of the invention include medical and veterinary subjects generally, including human patients. Among other subjects for whom the methods of the invention is useful are cats, dogs, large animals, avians such as chickens, and the like. In general, any subject who has a WBC deficiency or, more generally, who would profit from the elevation of white blood cell count is appropriate for administration of the invention method.

Typical conditions which are ameliorated or otherwise benefited by the method of the invention include hematopoietic disorders, such as aplastic anemia, leukemias, drug-induced anemias, and hematopoietic deficits from chemotherapy or radiation therapy. The method of the invention is also useful in enhancing the success of transplantation during and following immunosuppressive treatments as well as in

effecting more efficient wound healing and treatment of bacterial inflammation. The method of the present invention is further useful for treating subjects who are immunocompromised or whose immune system is otherwise impaired. Typical conditions which are ameliorated or otherwise benefited by the method of the present invention, include those subjects who are infected with a retrovirus and more specifically who are infected with human immunodeficiency virus (HIV). The method of the invention thus targets a broad spectrum of conditions characterized by a deficiency in white blood cell count, or which would benefit from elevation of said WBC count.

Having now generally described the invention, the same will be more readily understood through reference to the following examples which are provided by way of illustration, and are not intended to be limiting of the present invention, unless specified.

Example 1

Clinical Elevation of WBC Levels - Healthy Volunteers

Eleven human patients having initial white blood cell counts of 4,000-6,500 cells/mm^3 were used in the study. An intravenous dosing solution of AMD3100 (i.e., 1,1'-[1,4-phenylene-bis(methylene)]-bis-1,4,8,11-tetraazacyclotetradecane) were prepared from a stock solution which is a 1 mg/ml 1:10 dilution of a concentrate in 0.9% saline (normal saline) under sterile conditions. Aliquots from this stock solution were added to 50-ml bags of 0.9% saline for intravenous injection in amounts to achieve the desired dosage levels (10 μg/kg-80 μg/kg).

The subjects described in this example already contained an indwelling peripheral intravenous catheter. The prescribed amount of AMD3100 was administered over 15 minutes by intravenous fusion in a single dose. Blood samples were obtained prior to the dose, and at various times up to 24 hours after dose administration.

Eleven human subjects received intravenous administration of AMD-3100 at doses 10, 20, 40, and 80 μg/kg. Five subjects also received a single subcutaneous

injection of AMD-3100 at doses of 40 and 80 μg/kg. The effect of AMD3100 given intravenously in these 11 human subject is shown in Figure 1. Three patients were administered dosages of 10 μg/kg (open circles); 3 patients were administered dosages of 20 μg/kg (solid circles); 3 patients were administered 40 μg/kg (open triangles); and 2 patients were administered 80 μg/kg (closed triangles).

As shown in Figure 1, all of the patients at all levels of administration showed a marked increase in white blood cell count over the succeeding 5-10 hours after administration which WBC count tapered off after about 24 hours, although not, in any case, returning to the original level. Generally, the levels of WBC correlate with the concentration levels of the compound in the bloodstream. For example, one patient who received 80 μg/kg experienced an enhancement of white blood cell count from 6,000 cells/mm^3 to a peak value of 19,000 cells/mm^3. Even the patient showing the least response, who was given 20 μg/kg, experienced an increase from about 6,300 cells/mm^3 to about 9,000 cells/mm^3.

Thus, it appears that AMD3100 is consistently able to enhance WBC count in human patients.

While not intending to be bound by any theory, the ability to enhance WBC count across various species and the use of various compounds of formula (1) is believed due to the similarity of action of this compound in its antiviral applications and a possible mechanism for enhancing WBC count. The compounds of the invention are believed to exert their antiviral effects by inhibiting the binding of the second receptor for the HIV virus, CXCR-4, and thus to inhibit entry of the virus into the cell. These particular receptors appear homologous throughout a wide range of species, including mouse, rat, cat and man.

Example 2

Clinical Elevation of WBC Levels - HIV-Infected Patients

Elevations in WBC counts have also been observed in HIV-infected patients who received AMD-3100 by continuous infusion for up to 10 consecutive days (Figure 2). Eight patients received AMD-3100 at infusion dose rates of 2.5 μg/kg/hr

(patients 1-4) and 5.0 μg/kg/hr (patients 5-8). Elevations relative to the baseline were noted in samples taken on days 2, 6, and 11 (immediately prior to end of infusion) of the infusion period. Elevations in WBC count ratios (Day 11 samples) ranged from 1.4 to 2.8 times the baseline. WBC counts returned to baseline 7 days after discontinuation of the infusion. Thus, it appears that AMD3100 is consistently able to enhance WBC count following single dose or with continuous infusion in human patients.

While not intending to be bound by any theory, the ability to enhance WBC count across various species and the use of various compounds of formula (1) is believed due to the similarity of action of this compound in its antiviral applications and a possible mechanism for enhancing WBC count. The compounds of the invention are believed to exert their antiviral effects by inhibiting the binding of the second receptor for the HIV virus, CXCR-4, and thus to inhibit entry of the virus into the cell. These particular receptors appear homologous throughout a wide range of species, including mouse, rat, cat and man.

Claims

1. A method to treat a subject who would be benefited by elevation of white blood cell (WBC) count which method comprises

administering to said subject an amount of a compound of the formula

Z-linker-Z' (1)

or pharmaceutically acceptable salt thereof

wherein Z is a cyclic polyamine containing 9-32 ring members of which 3-8 are nitrogen atoms, said nitrogen atoms separated from each other by at least 2 carbon atoms, and wherein said heterocycle may optionally contain additional heteroatoms besides nitrogen and/or may be fused to an additional ring system;

Z' may be embodied in a form as defined by Z above, or alternatively may be of the formula

$$-N(R)-(CR_2)_n-X$$

wherein each R is independently H or straight, branched or cyclic alkyl (1-6C), n is 1 or 2, and X is an aromatic ring, including heteroaromatic rings, or is a mercaptan;

"linker" represents a bond, alkylene (1-6C) or may comprise aryl, fused aryl, oxygen atoms contained in an alkylene chain, or may contain keto groups or nitrogen or sulfur atoms;

in an amount effective to elevate said WBC count in said subject.

2. The method of claim 1 wherein Z and Z' are both cyclic polyamines.

3. The method of claim 1 wherein Z and Z' are identical.

4. The method of claim 1 wherein Z contains 10-24 members and contains 4 nitrogen atoms.

5. The method of claim 1 wherein Z and Z' are both 1,4,8,11-tetraazocyclotetradecane.

6. The method of claim 1 wherein the linker comprises an aromatic ring bracketed by two methylene moieties.

7. The method of claim 6 wherein the linker is 1,4-phenylene-bis-methylene.

8. The method of claim 7 wherein the compound of formula (1) is 1,1'-[1,4-phenylene-bis-(methylene)-bis-1,4,8,11-tetraazacyclotetradecane (AMD3100).

9. The method of claim 1 wherein the compound of formula (1) is

N-[1,4,8,11-tetraazacyclotetradecanyl-1,4-phenylenebis(methylene)]-2-aminomethyl)pyridine;

7,7'-[1,4-phenylenebis(methylene)]bis-4,7,10,17-tetraazabicyclo-[13.3.1] heptadeca-1(17),13,15-triene;

7,7'-[1,4-phenylenebis(methylene)]bis-3,7,11,17-tetraazabicyclo[13.3.1] heptadeca-1(17),13,15-triene;

1,1'-[1,3-phenylenebis(methylene)]-bis-1,4,8,11-tetra-azacyclotetradecane;

1,1'-[1,4-phenylenebis(methylene)]-bis-1,4,8,11-tetra-azacyclotetradecane;

1,1'-[1,4-phenylene-bis-(methylene)]-bis-1,4,7,10-tetraazacyclotetradecane;

1,1'-[1,3-phenylene-bis-(methylene)]-bis-1,4,7,10-tetraazacyclotetradecane;

11,11'-(1,2-propanediyl)bis-1,4,8,11-tetraazacyclotetradecane;

N-[4-(1,4,7-triazacyclotetra-decane)-1,4-phenylenebis(methylene)]-2-(aminomethyl)pyridine;

N-[7-(4,7,10-triazabicyclo[13.3.1]heptadeca-1(17),13,15-triene)-1,4-phenylenebis(methylene)]-2-(aminomethyl)pyridine;

N-[7-(4,7,10,17-tetraazabicyclo[13.3.1]heptadeca-1(17),13,15-triene)-1,4-phenylenebis(methylene)]-2-(aminomethyl)pyridine; or

N-[4-[4,7,10,17-tetraazabicyclo[13.3.1]heptadeca-1(17),13,15-triene]-1,4-phenylenebis(methylene)]-2-(aminomethyl)pyridine.

10. The method of claim 1 wherein formula (1) is in the form of its acid addition salt.

11. The method of claim 10 wherein the acid addition salt is the hydrobromide.

12. The method of claim 1 wherein the subject exhibits a hematopoietic deficit from chemotherapy or radiation therapy.

13. The method of claim 1 wherein the subject has a condition selected from the group consisting of aplastic anemia, leukemia and drug-induced anemia.

14. The method of claim 1 wherein the subject is a transplantation recipient.

15. The method of claim 1 wherein said elevation of WBC count enhances wound healing.

16. The method of claim 1 wherein said elevation of WBC count ameliorates bacterial inflammation.

17. The method of claim 1 wherein the compound is administered to said subject by an intravenous or subcutaneous route.

18. The method of claim 1 wherein the compound of formula (1) is administered to said subject in the dosage range of about 0.1 μg/kg-5 mg/kg of body weight.

19. A pharmaceutical composition comprising an effective amount of the compound of formula (1) as set forth in claim 1 in unit dosage form for elevating white blood cell count in a subject.

20. A method to stimulate the production of white blood cells (WBC) in an animal subject thereby elevating the WBC count which method comprises:

administering to said subject an amount of a compound of the formula

Z-linker-Z' (1)

or pharmaceutically acceptable salt thereof

wherein Z is a cyclic polyamine containing 9-32 ring members of which 3-8 are nitrogen atoms, said nitrogen atoms separated from each other by at least 2 carbon atoms, and wherein said heterocycle may optionally contain additional heteroatoms besides nitrogen and/or may be fused to an additional ring system;

Z' may be embodied in a form as defined by Z above, or alternatively may be of the formula

$$-N(R)-(CR_2)_n-X$$

wherein each R is independently H or straight, branched or cyclic alkyl (1-6C), n is 1 or 2, and X is an aromatic ring, including heteroaromatic rings, or is a mercaptan;

"linker" represents a bond, alkylene (1-6C) or may comprise aryl, fused aryl, oxygen atoms contained in an alkylene chain, or may contain keto groups or nitrogen or sulfur atoms;

in an amount effective to elevate said WBC count in said subject.

21. The method of claim 1 or claim 20, wherein said subject is infected with a retrovirus.

22. The method of claim 21, wherein said retrovirus is HIV.

23. The method of claim 1 or claim 20, wherein administering the compound of formula (1) to said subject is in a single dose or by continuous infusion.

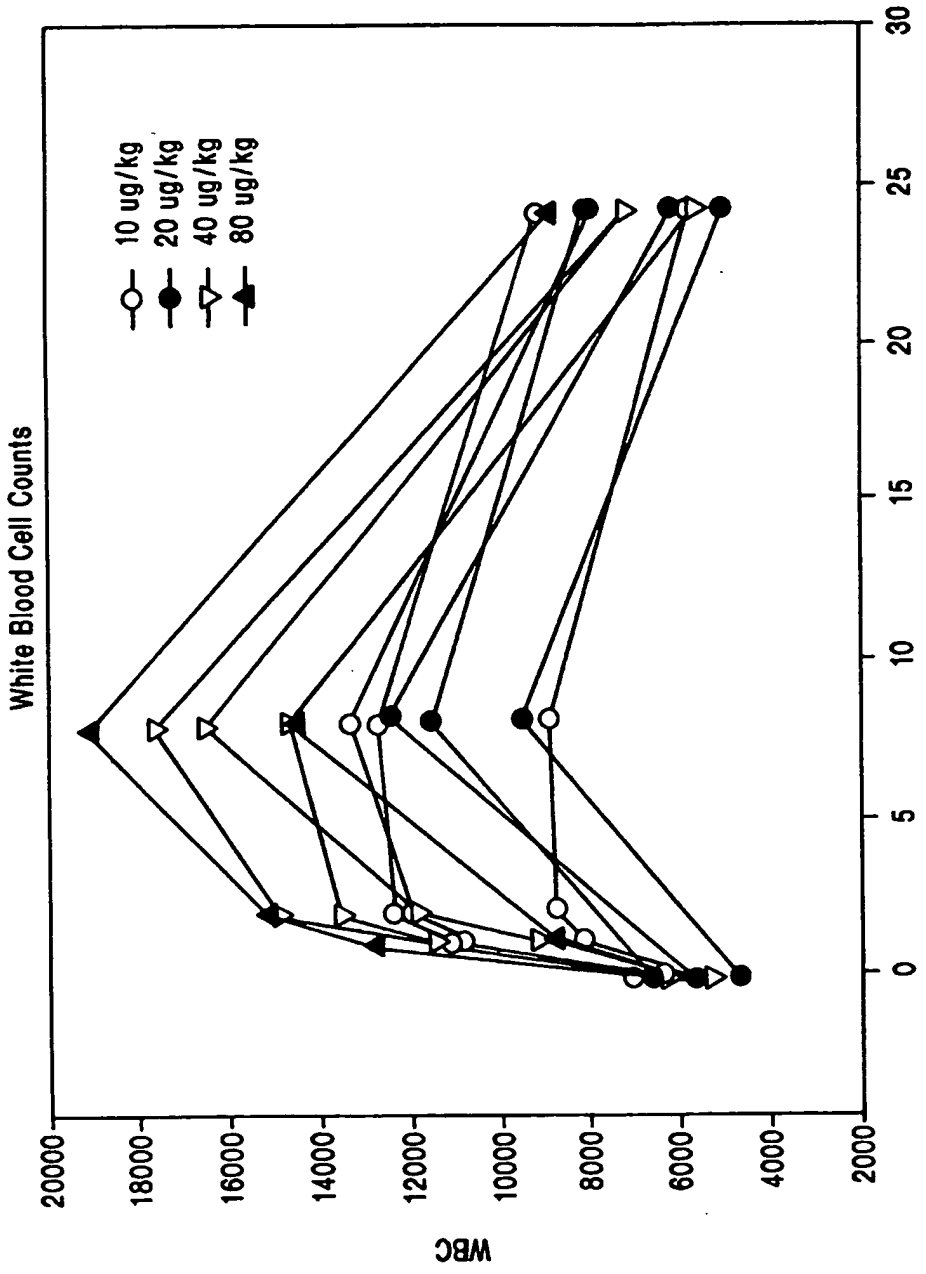
White Blood Cell Counts
10 ug/kg
20 ug/kg
40 ug/kg
80 ug/kg
WBC
20000
18000
16000
14000
12000
10000
8000
6000
4000
2000
Hours
0
5
10
15
20
25
30

FIG. 1

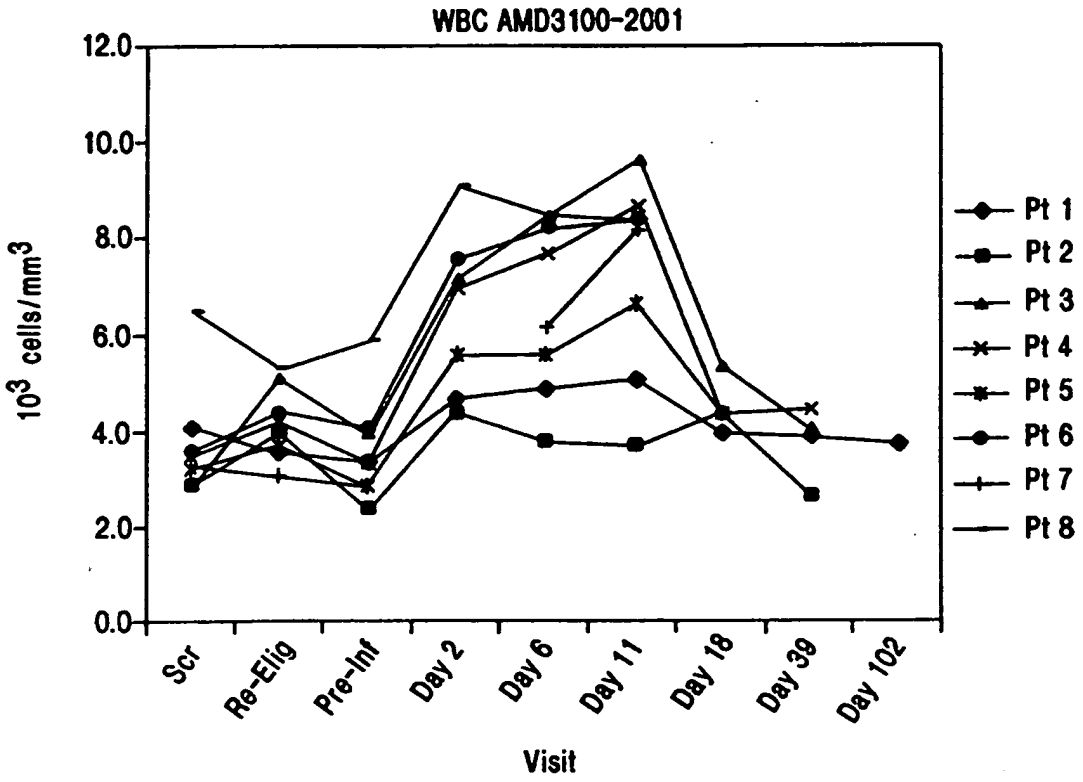
WBC AMD3100-2001
12.0
10.0
8.0
6.0
4.0
2.0
0.0
10^3 cells/mm^3
Scr
Re-Elig
Pre-Inf
Day 2
Day 6
Day 11
Day 18
Day 39
Day 102
Visit
Pt 1
Pt 2
Pt 3
Pt 4
Pt 5
Pt 6
Pt 7
Pt 8

FIG. 2

INTERNATIONAL SEARCH REPORT

International Application No
PCT/CA 00/00104

A. CLASSIFICATION OF SUBJECT MATTER
IPC 7 A61K31/395 A61K31/4427

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
IPC 7 A61K

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)
EPO-Internal

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
P,X	WO 00 02870 A (SCHOLS DOMINIQUE ;ANORMED INC (CA); WANG ZHONGREN (CA); BOEHRINGER) 20 January 2000 (2000-01-20) cited in the application page 9, line 12-27 page 9, line 30 -page 11, line 7 claims 21,31; figures 1-28; examples ---	1,4,5,7, 9-12,14, 17-23
X	WO 95 18808 A (JOHNSON MATTHEY PLC ;BRIDGER GARY JAMES (US); PADMANABHAN SREENIVA) 13 July 1995 (1995-07-13) cited in the application page 1, line 1 -page 4, line 4 * See compound E * page 27-28; claims ---	1-4,6,7, 9-11, 17-23
	-/--	

[X] Further documents are listed in the continuation of box C.

[X] Patent family members are listed in annex.

° Special categories of cited documents :

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier document but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.

"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
17 July 2000	25/07/2000

Name and mailing address of the ISA	Authorized officer
European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Tx. 31 651 epo nl, Fax: (+31-70) 340-3016	Veronese, A

Form PCT/ISA/210 (second sheet) (July 1992)

INTERNATIONAL SEARCH REPORT

International Application No
PCT/CA 00/00104

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	EP 0 434 385 A (JOHNSON MATTHEY PLC) 26 June 1991 (1991-06-26) page 2, line 1-26 page 5, line 34 -page 6, line 14 page 6, line 26; claims	1-5,10, 11,15, 17-23
X	BRIDGER G J ET AL: "SYNTHESIS AND STRUCTURE-ACTIVITY RELATIONSHIPS OF PHENYLENEBIS(METHYLENE)-LINKED BIS-TETRAAZAMACROCYCLES THAT INHIBIT HIV REPLICATION. EFFECTS OF MACROCYCLIC RING SIZE AND SUBSTITUENTS ON THE AROMATIC LINKER" JOURNAL OF MEDICINAL CHEMISTRY,US,AMERICAN CHEMICAL SOCIETY. WASHINGTON, vol. 38, no. 2, 20 January 1995 (1995-01-20), pages 366-378, XP000196379 ISSN: 0022-2623 the whole document	1-11, 19-23
A	US 5 582 823 A (SOUZA LAWRENCE M) 10 December 1996 (1996-12-10) cited in the application the whole document	1-23
A	HUNT D W C ET AL: "PHOTOFRIN, BUT NOT BENZOPORPHYRIN DERIVATIVE, STIMULATES HEMATOPOIESIS IN THE MOUSE" IMMUNOPHARMACOLOGY,XX,ELSEVIER SCIENCE PUBLISHERS BV, vol. 26, no. 3, 1 November 1993 (1993-11-01), pages 203-212, XP000567096 ISSN: 0162-3109 the whole document	1-23

1

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

Continuation of Box I.2

The definition "method to treat a subject who would be benefited by elevation of white blood cell count" is not defining a specific disease recognized in the art to which the invention belongs. This definition relates to all diseases which could benefit by elevation of white blood cells count, whereas the application provides support within the meaning of Article 6 PCT and/or disclosure within the meaning of Article 5 PCT for only a limited number of such diseases.
Moreover, the definition of Markush formula in claim 1-7 relates to an extremely large number of possible compounds. Also in this case, the application provides support within the meaning of Article 6 PCT and/or disclosure within the meaning of Article 5 PCT for only a very limited number of such compounds.
In the present application, the above mentioned extremely large number of compounds is claimed for the treatment of a number of conditions in a subject who would be benefited by elevation of white blood cell count: (haematopoietic deficit, anaemia, leukemia, transplantation, wound healing, bacterial infections and others). A direct relation between the mechanism of action which has been discovered, and the very broad spectrum of diseases mentioned in the application, is however not demonstrated (Art. 6 PCT).
Moreover, the use of some of the compounds falling under the above mentioned definition is already known in relation to the treatment of the same diseases mentioned by the inventors.
It is clear that a meaningful search of the whole of the claimed scope is virtually impossible in this situation, because of the extremely large number of compounds defined and because of the large number of diseases allegedly related to the above mentioned pharmacological mechanism.
In the present application support within the meaning of Article 6 PCT and/or disclosure within the meaning of Article 5 PCT is to be found only for a very small proportion of the compounds and the diseases. In the present case, the claims so lack support, and the application so lacks disclosure, that a meaningful search over the whole of the claimed scope is impossible.

Consequently, the search has been carried out for those parts of the claims which appear to be supported and disclosed, namely the compounds disclosed in the examples and in claims 8,9 in relation to the treatment of the diseases mentioned in claims12,13,14,15,16, 22.

Claims searched incompletely: 1-23 (claims 8,9 have been searched incompletely in respect to the diseases for which the compounds are meant to be useful).

The applicant's attention is drawn to the fact that claims, or parts of claims, relating to inventions in respect of which no international search report has been established need not be the subject of an international preliminary examination (Rule 66.1(e) PCT). The applicant is advised that the EPO policy when acting as an International Preliminary Examining Authority is normally not to carry out a preliminary examination on matter which has not been searched. This is

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

the case irrespective of whether or not the claims are amended following receipt of the search report or during any Chapter II procedure.

INTERNATIONAL SEARCH REPORT

Information on patent family members

International Application No
PCT/CA 00/00104

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